Separation and Determination of Aliphatic Amines in Environmental Matrices by CE Using Indirect UV and Laser-Induced Fluorescence Detection

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EXPERIMENTAL

CE experiments were carried out using a Beckman P/ACE 2100 or 5000 instrument with indirect UV detection or laser-induced fluorescence detection (Ar ion laser, 488 nm excitation, 520/20 nm emission).

Solid phase extraction (SPE) studies used SCX cartridges, C18 extraction disks with ion-pairing, or cation exchange extraction disks. Elution was 4% ammonia in methanol.

Amines were derivatized using fluorescein isothiocyanate at 55º C with sodium bicarbonate buffer, usually in aqueous solution. An alternate solvent was acetonitrile. CE separations using indirect detection under free zone conditions used imidazole at 5 mM at pH 5.0 or as otherwise indicated in the tables.

Separations under LIF detection were free zone at pH 7.0 of 5:1 buffer (50 mM phosphate:methanol). Separations under MEKC were at pH 7.0 using urea, SDS, and methanol as indicated on figure.

BACKGROUND

Aliphatic amines are toxic substances and irritants to mucous membranes that are among the common chemicals of commerce. They are used as corrosion inhibitors in steam boilers and as starting materials in the manufacture of pharmaceuticals, insecticides, herbicides, fungicides, polymers, surfactants, and rubber accelerators. The related alkanolamines function as solvents and starting materials for surfactants, but they appear to be less toxic than the aliphatic amines.

The many commercial uses and natural occurrence of aliphatic amines (here we refer primarily to C_1 to C_4 alkyl substituted primary, secondary, and tertiary amines) suggest that ultimately they will appear in the environment as pollutants. Thus, they are target analytes of U.S. EPA Method 8260 (also Method 624) where they are classified as volatiles. The U.S. EPA, EMSL-LV, maintains a continuing interest in analytical methods for amines because of their wide occurrence. In addition, there is need for determinative methods for amines as a result of the listing activities for various hazardous wastes under RCRA when amines are suspected to be present.

RESULTS

compound	effective mobility (10^{-4} cm^2 V^{-1}s^{-1})		
	imidazole (5.0 *mM*, pH 5.0)	N-ethylbenzylamine (5.5 *mM*, pH 5.1)	benzyltriethylammonium chloride (5.0 *mM*, pH 10.1)
tetrabutylammonium bromide	1.81	1.81	1.75
triethanolamine	2.83	N/A	1.75
dipropylamine	2.76	N/A	2.53
tripropylamine	2.35	2.35	2.28
dibutylamine	2.42	2.42	2.29
butylamine	3.33	N/A	2.23
diethylamine	3.37	N/A	2.61

1 **Slopes (relative response per u*M*) are based on 5-10 determinations for each compound and were taken from plots like those presented in Figure 2. Linear regression analyses were performed on each data set. Values for R^2 exceeded 0.99 in each case. For other methods, see Figure 1 and text.**

2 **Effective mobilities (10^{-4} cm^2 V^{-1}s^{-1}) were calculated from measured migration times by the following equation: u_a = apparent mobility; u_e = effective mobility; u_{EOF} = mobility provided by electroosmotic flow.**

$$u_a = \ell L / tV$$

where

u_a	=	$u_e + u_{EOF}$
V	=	applied voltage
ℓ	=	effective capillary length (to detector)
t	=	migration time
L	=	total capillary length

Table 2.
Solid Phase Extraction of Propylamines from Aqueous Solution: Sodium Dodecylbenzenesulfonate as Ion Pairing Agent

	percent recovery					
	propy-lamine		dipropy-lamine		tripropy-lamine	
sample[1]	Exp.1	Exp.2	Exp.1	Exp.2	Exp.1	Exp.21
	70	70	77	77	61	61
2	86	86	88	88	65	65
3	92	92	103	100	70	70
Average	82		89		65	

1 **Samples 1, 2, and 3 consisted of 300 μmol of dodecylbenzenesulfonate (Na$^+$); 37 μmol propylamine; 33 μmol dipropylamine; 31 μmol tripropylamine; 5 mL methanol in a final volume of 1 L. Each of the three samples was passed through a separate disk prepared as suggested by the manufacturer (Empore Bakerbond Octadecyl C_{18}). The 100% level of recovery was established by an appropriate portion of the original solutions used in preparation of the samples.**

Table 3.
Recovery of Amines from a More Dilute Aqueous Solution

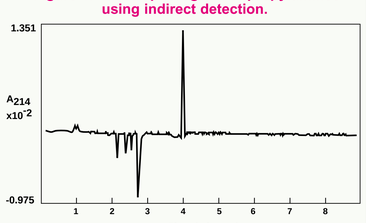
	percent recovery[2]		
disk[1]	propylamine	dipropylamine	tripropy-lamine
1	105	107	108
2	98	112	107
3	96	96	86
4	0	0	0

1 **Empore Bakerbond Octadecyl (C_{18}) extraction disks were prepared following the manufacturer's instructions.**

2 **The test solution contained per L, 45 μmol of total amines; 150 μmol of dodecylbenzene-sulfonate, sodium salt; and 5.0 mL of methanol. The filtrate from disk 3 was passed through disk 4 to check for amines remaining in solution after the first passage.**

INDIRECT UV

Figure 1. Electropherogram of propylamines using indirect detection.



Compound	Migration Time Seconds	Mobility μ_e 10^{-4}cm^{2}V^{-1}S^{-1}	Response Factor Per mM	10^4 Theoretical Plates
propylamine	119	3.76	0.385	5.48
dipropylamine	135	2.83	0.421	3.13
tripropylamine	144	2.39	0.395	5.55
tetrabutylammonium bromide	155	1.81	-	0.75

Figure 2. Response is a linear function of concentration.

Compound	Lowest Detected Concentration mMolar	Mass Detection Limit 10^{-4} nMoles
Propylamine	0.033	1.65
Dipropylamine	0.020	1.0
Tripropylamine	0.014	0.7
Ethanolamine	0.039	1.9
Diethanolamine	0.026	1.3
Triethanolamine	0.018	0.9

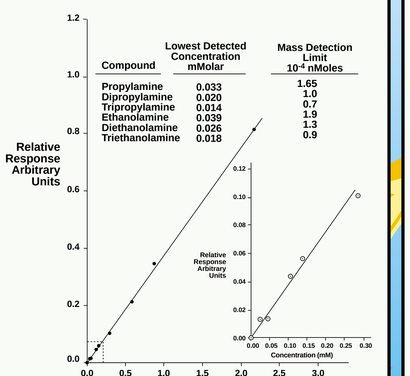


Table 4.
Solid Phase Extraction of Organic Amines from Aqueous Solution: Sulfonic Acid Bonded to Poly (styrenedivinylbenzene) as Cationic Exchange Resin.[1]

	percent recovery			
compound[2]	sample 1	sample 2	sample 3	average
butylamine	49	67	60	59
dibutylamine	50	66	76	64
tributylamine	42	58	46	49
dimethylamine	12	15	23	17
diethylamine	10	14	18	14
triethylamine	12	16	20	16
propylamine	79	85	83	82
dipropylamine	72	84	78	78

1 **Cation exchange disks (47-mm, sulfonic acid bonded to poly (styrenedivinylbenzene) copolymer, hydrogen form) were a gift of 3M, new products department, St. Paul, MN 55144. Disks were prepared by washing with acetone, methanol, water, very dilute H_2SO_4 (2 drops/100 mL), and finally with H_2O until the pH reached 5.5. Disks were eluted with four 5.0-mL portions of 4% NH_4OH in methanol (v/v); following elution, the disks were regenerated as indicated in the washing procedure.**

2 **Samples consisting of 1L were passed through the disk at a flow rate of approximately 200 mL/min.**

Figure 3. Mobilities of amines are correlated with 1/Stokes radius.

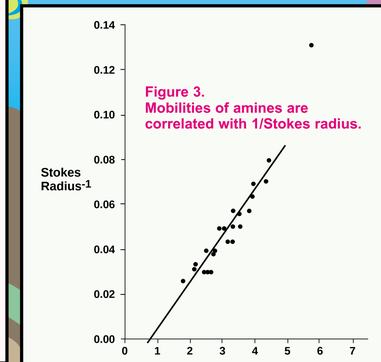
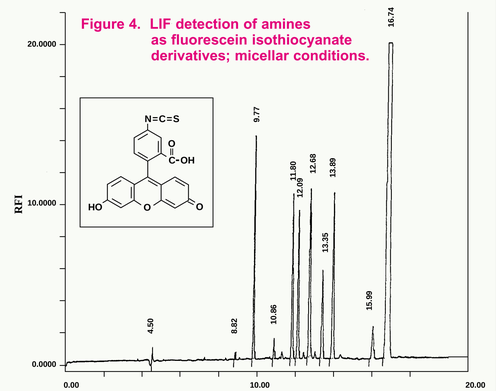


Figure 4. LIF detection of amines as fluorescein isothiocyanate derivatives; micellar conditions.



Separation of aliphatic and other amines with LIF detection; buffer and conditions: phosphate (0.039 M), SDS (0.070 M), urea (2.0 M), 23 methanol, pH 7.0, 47 cm X 0.050 mm ID capillary, 30 kV, LIF detection (488 nm excitation and 520/20 nm emission, 40 cm to detector). Peak identities (as fluorescein isothiocyanate derivatives): MT =9.77, 2-(2-aminoethyl)pyridine; MT =11.80, 1,5-diaminopentane; MT =12.09, p-toluidine; MT =12.68, butylamine; MT =13.35, diethylamine; MT =13.89, propylamine.

Figure 5. LIF detection of amines under free zone separation; 1,5-diaminopentane is well separated.

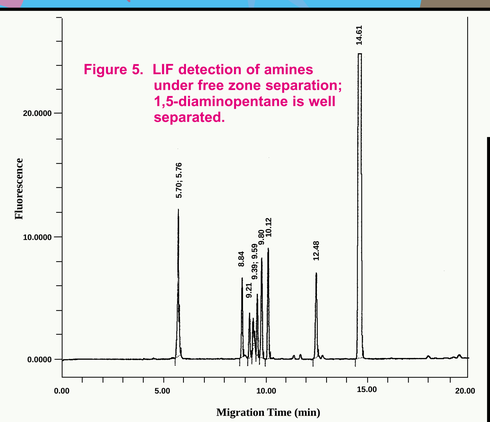
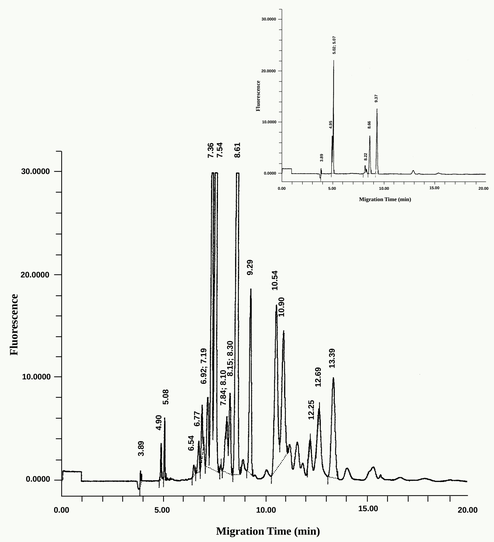


Figure 6. Low ng (ppb) level derivatization of 1,5-aminopentane with LIF detection; background problems.



CONCLUSION

The aliphatic amines may be determined in aqueous matrices by indirect methods provided the background interferences are minimal. SPE may be used to afford 100 to 1000 fold concentrations of selected analytes. Electrophoretic mobilities were correlated with the Stokes radius of each analyte ion. Responses were a linear function of concentration. Separations of closely migrating ions was effected by use of optimal background electrolyte, additives, or pH adjustment to the pK_b of the amines.

LIF detection offers improved sensitivity and robustness with respect to inorganic interferences. Limitations include derivatization of matrix coextractives and by-products of derivatization for ng and sub-ng amounts of amines. Two orthogonal chromatographies are being investigated to address these limitations. GPC provides some removal of many matrix coextractives prior to derivatization. Peak selection from reverse-phase HPLC of the fluorescein isocyante derivatives provides a cleanup step prior to final separation/determination by CZE or MEKC. Derivatization of tertiary amines is under investigation by other reagents.